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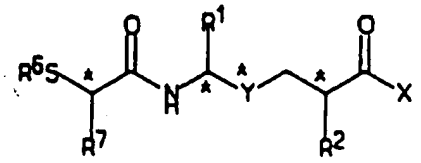
(54) Title: MERCAPTOAMIDE DERIVATIVES AND THEIR THERAPEUTIC USE

(57) Abstract

Mercaptoamide derivatives of formula (I), in which Y represents CHOH, $CHNH_2$ or C=O and the other variables are defined in the description, have therapeutic activity as metalloproteinase, TNFalpha and L-selectin sheddase inhibitors.



(I)

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MERCAPTOAMIDE DERIVATIVES AND THEIR THERAPEUTIC USE

Field of the Invention

This invention relates to a novel class of peptidyl derivatives, to processes for their preparation, and to their use in medicine.

Background to the Invention

Metalloproteinases, (human fibroblast) collagenase, stromelysin, gelatinase, tumour necrosis factor (TNF), L-selectin sheddase and their modes of action, and also inhibitors thereof and their clinical effects, are disclosed in WO-A-9611209 and our other PCT Application having the same title and filing date, the contents of which are incorporated by reference.

In normal tissues, cellular connective tissue synthesis is offset by extracellular matrix degradation, the two opposing effects existing in dynamic equilibrium. Degradation of the matrix is brought about by the action of proteinases released from resident connective tissue cells and invading inflammatory cells, and is due, in part, to the activity of at least three groups of metalloproteinases. These are the collagenases (interstitial collagenase, MMP-1; PMN collagenase, MMP-8; collagenase-3, MMP-13), the gelatinases (gelatinase A, MMP-2, 72kDa-gelatinase, Type IV collagenase; gelatinase B, MMP-9, 92kDa-gelatinase, Type IV collagenase) and the stromelysins (proteoglycanase, MMP-3, stromelysin-1, transin; stromelysin-2, MMP-10; stromelysin 3, MMP-11). Normally these catabolic enzymes are tightly regulated at the level of their synthesis and secretion and also at the level of their extracellular activity, the latter through the action of specific inhibitors, such as TIMP (tissue inhibitors of metalloproteinase), which form inactive complexes with metalloproteinases, and more general proteinase inhibitors such as α_2-macroglobulins.

The accelerated, uncontrolled breakdown of connective tissues by metalloproteinase catalysed resorption of the extracellular matrix is a feature of many pathological conditions such as rheumatoid arthritis, osteoarthritis, septic arthritis, corneal, epidermal or gastric ulceration; tumour metastasis or invasion; periodontal disease, proteinuria, coronary thrombosis associated with atherosclerotic plaque rupture and bone disease. The inhibitors claimed herein may also be useful in preventing the pathological squaelae following a traumatic injury that could lead to a permanent disability. These compounds may also have utility as a means for birth control by preventing ovulation or implantation. It can be expected that the pathogenesis of such diseases is likely to be modified in a beneficial manner by the administration of metalloproteinase inhibitors and numerous compounds have been suggested for this purpose [for a general review see R C Wahl, *et al* Ann. Rep, Med. Chem. 25: 175-184, Academic Press Inc., San Diego (1990)].

A number of small peptide like compounds which inhibit metalloproteinases have been described. Perhaps the most notable of these are those relating to angiotensin converting enzyme (ACE) where such agents act to block the conversion of the decapeptide angiotensin I to angiotensin II, a potent pressor substance. Compounds of this type are described in EP-A-0012401. Also, related mercaptoamide peptidyl derivatives have shown ACE inhibitor activity *in vitro* and *in vivo* (H N Weller *et al* (1984), Biochem Biophys. Res. Comm., 125 (1):82-89).

TNFα is a cytokine which is produced initially as a cell-associated 28kD precursor. It is released as an active, 17kD form (D-M Jue *et al*, (1990) Biochemistry, 29:8371-8377), which can mediate a large number of deleterious effects *in vivo*. When administered to animals or humans it causes inflammation, fever, cardiovascular effects, haemorrhage, coagulation and acute phase responses,

similar to those seen during acute infections and shock states. Chronic administration can also cause cachexia and anorexia. Accumulation of excessive TNFα can be lethal.

There is considerable evidence from animal model studies that blocking the effects of TNFα with specific antibodies can be beneficial in acute infections, shock states, graft versus host reactions and autoimmune disease. TNFα is also an autocrine growth factor for some myelomas and lymphomas and can act to inhibit normal heamatopoiesis in patients with these tumours.

Preventing the production or action of TNFα is, therefore, predicted to be a potent therapeutic strategy for many inflammatory, infectious, immunological or malignant diseases. These include, but are not restricted to, septic shock, haemodynamic shock and sepsis syndrome (Mathison *et al* (1988) J. Clin. Invest. 81:1925-1937; Miethke *et al* (1992), J. Exp. Med. 175:91-98), post ischaemic reperfusion injury, malaria (Grau *et al* (1989), Immunol. Rev. 112:49-70); mycobacterial infection (Barnes *et al* (1992) Infect. Imm. 60:1441-6), meningitis, psoriasis, congestive heart failure, fibrotic disease, cachexia, graft rejection, cancer, autoimmune disease, rheumatoid arthritis, multiple sclerosis, radiation damage, toxicity following administration of immunosuppressive monoclonal antibodies such as OKT3 or CAMPATH-1 and hyperoxic alveolar injury.

Current clinical anti-TNFα strategies involve the use of corticosteroids such as dexamethasone, and the use of cyclosporin-A or FK506, which are non-specific inhibitors of cytokine gene transcription. Phosphodiesterase inhibitors such as pentoxyfilline have been shown to be more specific inhibitors of TNFα gene transcription (Endres S . (1991) Immunol. 72:56-60, Schandene *et al* (1992), Immunol. 76:30-34, Alegre ML, *et al* (1991); Transplantation 52:674-679, Bianco *et al* (1991) Blood 78:1205-1221). Thalidomide has also been shown to inhibit TNFα production by leucocytes (Sampajo *et al* (1991), J. Exp. Med. 173:699-703). In experimental settings, anti-TNFα monoclonal antibodies, soluble TNF receptors and soluble TNFα receptor/immunoadhesins have been shown to specifically inhibit the effects of TNFα action (Bagby *et al* (1991) J. Infect. Dis. 163:83-88, Charpentier *et al*. (1991) Presse-med. 20:2009-2011, Silva *et al* (1990) J. Infect. Dis. 162:421-427; Franks *et al* (1991) Infect. Immun. 59:2609-2614, Tracey *et al* (1987) Nature 330:662-664; Fischer *et al* (1992) PNAS USA in press, Lesslauer *et al* (1991) Eur. J. Immunol. 21:2883-2886, Ashkenazi *et al* (1991) PNAS USA 88:10535-10539).

It has recently been shown that the effects of TNF are mediated by two peptides, TNFα and TNFβ. Although these peptides have only 30% homology with each other, they activate the same receptors and are encoded by immediately adjacent genes. As used herein, the term tumour necrosis factor or TNF therefore means tumour necrosis factor a and peptides having a high degrees of sequence homology with, or substantially similar physiological effects to, TNFα, for example TNFβ.

One of the objectives of the present invention is to provide compounds which substantially inhibit the release of TNF from cells, and therefore may be used in the treatment of conditions mediated by TNF. Such uses include, but are not limited to, the treatment of inflammation, fever, cardiovascular effects, haemorrhage, coagulation and acute phase response, cachexia and anorexia, acute infections, shock states, graft versus host reactions and autoimmune disease.

Compounds which have the property of inhibiting the action of metalloproteinases involved in connective tissue breakdown such as collagenase, stromelysin and gelatinase have been shown to inhibit the release of TNF both *in vitro* and *in vivo* (AJH Gearing *et al* (1994), Nature, 370:555-557; GM McGeehan *et al* (1994), Nature, 370:558-561: MJ Crimmin *et al*, WO 93/20047). All of these reported inhibitors contain a hydroxamic acid zinc binding group.

It is, therefore, a further objective of this invention to provide compounds which, in addition to inhibiting TNF release, also inhibit the action of MMPs, and hence may be used in the treatment of patients who suffer from conditions mediated by TNF and/or MMPs.

As appreciated by those of skill in the art the significant proportion of homology between human fibroblast collagenase, stromelysin and gelatinase leads to the possibility that a compound that inhibits one enzyme may to some degree inhibit all of them.

Compounds that inhibit collagenase, which possess structural portions akin to those of the instant invention include those encompassed by U.S.4,511,504 issued Apr. 16, 1985; U.S. 4,568,666, issued Feb 4, 1986.

Compounds of related structure that are claimed to inhibit stromelysin (proteoglycanase) are encompassed by U.S.4,771,037, issued Sept. 13, 1988.

The applicants believe that stromelysin and collagenase inhibitors have utility in preventing articular cartilage damage associated with septic arthritis. Bacterial infections of the joints can elicit an inflammatory response that may then be perpetuated beyond what is needed for removal of the infective agent resulting in permanent damage to structural components. Bacterial agents have been used in animal models to elicit an arthritic response with the appearance of proteolytic activities. See J. P. Case *et al* (1989), J. Clin. Invest., 84:1731-40; R. J. Williams *et al* (1990), Arth. Rheum., 33: 533-41.

The applicants also believe that inhibitors of stromelysin, collagenase and gelatinase will be useful to control tumour metastasis, optionally in combination with current chemotherapy and/or radiation. See L. M. Matrisian *et al* (1986), Proc. Natl. Acad. Sci., USA, 83:9413-7; S. M. Wilhelm *et al* (1987), Ibid. 84:6725-29; Z. Werb *et al* (1989), J. Cell Biol., 109:872-889; L. A. Liotta *et al* (1983), Lab. Invest., 49:636-649; R. Reich *et al* in Metatasis; Ciba Foundation Symposium, Wiley, Chicester, 1988, pp. 193-210.

Secreted proteinases such as stromelysin, collagenase and gelatinase play an important role in processes involved in the movement of cells during metastasic tumour invasion. Indeed, there is also evidence that the matrix metalloproteinases are overexpressed in certain metastatic tumour cell lines. In this context, the enzyme functions to penetrate underlying basement membranes and allow the tumour cell to escape from the site of primary tumour formation and enter the circulation. After adhering to blood vessel walls, the tumour cells use these same metalloproteinases to pierce underlying basement membranes and penetrate other tissues, thereby leading to tumour metastasis. Inhibition of this process would prevent metastasis and improve the efficacy of current treatments with chemotherapeutics and/or radiation.

These inhibitors should also be useful for controlling periodontal diseases, such as gingivitis. Both collagenase and stromelysin activities have been isolated from fibroblasts derived from inflamed gingiva (V. J. Uitto *et al* (1981), J.Periodontal Res., 16:417-424). Enzyme levels have been correlated to the severity of gum disease; C. M. Overall *et al* (1987), J. Periodontal Res., 22:81-88.

Proteolytic processes have also been observed in the ulceration of the c rnea following alkali burns (S. I. Brown *et al* (1969), Arch. Opthalmol., 81:370-373). Mercapto-containing peptides do inhibit the collagenase isolated from alkali-burned rabbit cornea (F. R. Burns *et al* (1989), Invest. Opthalmol, 30:1569-1575). Treatment of alkali-burned eyes or eyes exhibiting corneal ulceration as a result of infection with inhibitors of these metalloendoproteinases in combination with sodium

citrate or sodium ascorbate and/or antimicrobials may be effective in preventing developing corneal degradation.

Stromelysin has been implicated in the degradation of structural components of the glomerular basement membrane (GBM) of the kidney, the major function of which is to restrict passage of plasma proteins into the urine (W. H. Baricos *et al* (1989), Biochem. J., 254:609-612). Proteinuria, a result of glomerular disease, is excess protein in the urine caused by increased permeability of the GBM to plasma proteins. The underlying causes of the increased GBM permeability are unknown, but proteinases including stromelysin may play an important role in glomerular diseases. Inhibition of this enzyme may alleviate the proteinura associated with kidney malfunction.

It is suggested that inhibition of stromelysin activity may prevent the rupturing of atherosclerotic plaques leading to coronary thrombosis. The tearing or rupture of atherosclerotic plaques is the most common event initiating coronary thrombosis. Destabilisation and degradation of the connective tissue matrix surrounding these plaques by proteolytic enzymes or cytokines released by infiltrating inflammatory cells has been proposed as a cause of plaque fissuring. Such tearing of these plaques can cause an acute thrombolytic event as blood rapidly flows out of the blood vessel. High levels of stromelysin RNA message have been found to be localised to individual cells in atherosclerotic plaques removed from heart transplant patients at the time of surgery (A. M. Henney *et al* (1991), Proc. Nat'l. Acad. Sci. USA, 88:8154-8158). Inhibition of stromelysin by these compounds may aid in preventing or delaying the degradation of the connective tissue matrix that stabilises the atherosclerotic plaques, thereby preventing events leading to acute coronary thrombosis.

It is also believed that specific inhibitors of stromelysin and collagenase should be useful as birth control agents. There is evidence that expression of metalloproteinases, including stromelysin and collagenase, is observed in unfertilised eggs and zygotes and at further cleavage stages and increased at the blastocyst stage of fetal development and with endoderm differentiation (C. A. Brenner *et al* (1989), Genes & Develop., 3:848-59). By analogy to tumour invasion, a blastocyst may express metalloproteinases in order to penetrate the extracelluar matrix of the uterine wall during implantation. Inhibition of stromelysin and collagenase during these early development processes should presumably prevent normal embryonic development and/or implantation in the uterus. Such intervention would constitute a novel method of birth control. In addition there is evidence that collagenase is important in ovulation processes. In this example, a covering of collagen over the apical region of the follicle must be penetrated in order for the ovum to escape. Collagenase has been detected during this process and an inhibitor has been shown to be effective in preventing ovulation (J. F. Woessner *et al* (1989), Steroids, 54:491-499). There may also be a role for stromelysin activity during ovulation (C. K. L. Too *et al* (1984), Endocrin., 115:1043-1050).

Collagenolytic and stromelysin activity have also been observed in dystrophic epidermolysis bullosa (A. Kronberger *et al* (1982), J. Invest. Dermatol., 79:208-211; D. Sawamura *et al* (1991), Biochem. Biophys. Res. Commun., 184:1003-8). Inhibition of metalloendoproteinases should limit the rapid destruction of connective components of the skin.

In addition to extracelluar matrix comprising structural components, stromelysin can degrade other *in vivo* substrates including the inhibitors α_1-proteinase inhibitor and may therefore influence the activities of other proteinases such as elastase (P. G. Winyard *et al* (1991), FEBS Letts., 279,1:91-94). Inhibition of the matrix metalloendoproteinases may potentiate the antiproteinase activity of these endogenous inhibitors.

From recent publications it is evident that several new enzymes of the MMP family have been identified, some of which maybe important in disease. Collagenase 3, an enzyme unique to breast carcinoma cells may have utility in breast cancer (JMP Freije *et al* (1994), J. Biol. Chem., 269 (24): 16766-16773), whilst MT-MMP, another member of the MMP family has been shown to be a key enzyme in the activation of gelatinase A (H Sato *et al* (1994), Nature, 370:61-65). Gelatinase A is an important enzyme in the growth and metastasis of tumours (such as defined above).

The degradation of b-Amyloid Precusor Protein (APP) has been shown to generate amyloid plaques, a major constituent of the senile plaques, found in patients with Alzheimers Disease (AD). Two recent publications have identified metalloproteinase enzymes that cleave APP to the amyloid plaque (CR Abraham *et al* (1994), Biochemistry, 33:192-199; G Huber *et al* (1994), Biochem. Biophys. Res. Comm., 201 (1):45-53).

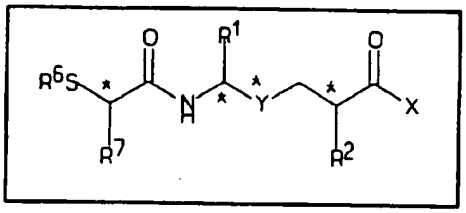
As appreciated by those of skill in the art, the significant proportion of homology between these new enzymes and other MMPs leads to the possibility that a compound that inhibits one enzyme may to some degree inhibit these new enzymes. Therefore, inhibitors encompassed in this invention may be useful in the diseases in which these new enzymes are implicated.

SUMMARY OF THE INVENTION

The invention encompasses novel mercaptoacyl compounds of formula (I) which are useful inhibitors of matrix metalloproteinase and/or TNF mediated diseases including degenerative diseases (such as defined above and in WO-A-9611209 etc.) and certain cancers.

In a first aspect of the invention there is provided a compound of general formula (I)

(I)



wherein:

R^1 is a C_{1-7} alkyl, C_{2-6} alkenyl, (C_{1-6} alkyl)aryl, (C_{1-6} alkyl)heteroaryl or C_{1-6} alkyl-AR^8 group where A is O, NR^8 or $S(O)_n$ where n = 0-2, and R^8 is H, C_{1-4} alkyl, aryl, heteroaryl, (C_{1-4} alkyl)aryl or (C_{1-4} alkyl)heteroaryl; if A = NR^8 the groups R^8 may be the same or different.

R^2 is a $[Alk]_mR^5$ group where Alk is a C_{1-6} alkyl or C_{2-6} alkenyl group and m = 0-1;

X is NR^3R^4 where R^3 is hydrogen or the group C_{1-6} alkyl optionally substituted by amino (NH_2), aryl, arylamino, protected amino, di (C_{1-6}alkyl)amino, mono (C_{1-6}alkyl)amino, CO_2H, protected carboxyl, carbamoyl, mono (C_{1-6}alkyl) carbamoyl, di (C_{1-6}alkyl) carbamoyl, and R^4 is hydrogen or a C_{1-6} alkyl group; NR^3R^4 may also form a 5-7 membered ring such as pyrrolidine, piperidine or morpholine;

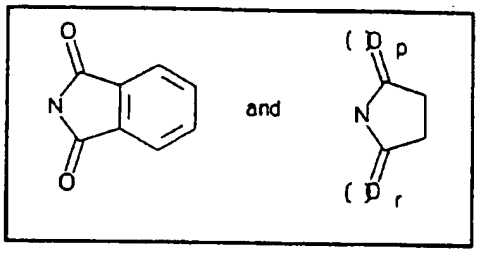
Y is CHOH, $CHNH_2$ or C=O;

R^5 is an optionally substituted cyclo(C_{3-6})alkyl, cyclo(C_{3-6})alkenyl, C_{1-6} alkyl, aryl, heteroaryl, CO_2R^8, $CONHR^8$, $NHCO_2R^8$, $NHSO_2R^8$, $NHCOR^8$ or AR^8 group;

R^6 is H or the group R^9CO where R^9 is a C_{1-4} alkyl or aryl group;

R^7 is aryl (optionally substituted with R^{10}), heteroaryl (optionally substituted with R^{10}), C_{1-4} alkyl (optionally substituted with R^{10}), C_{1-4} alkenyl (optionally substituted with R^{10}), (C_{1-4} alkyl)aryl (optionally substituted with R^{10}), (C_{1-4} alkyl)heteroaryl (optionally substituted with R^{10}), cyclo(C_{3-6})alkyl (optionally substituted with R^{10}) or cyclo(C_{3-6})alkenyl (optionally substituted with R^{10});

R^{10} is H, SR^6, COR^{11}, $N(R^8)_2$ (where R^8 may be the same or different), NR^8R^{12}, OR^8, COR^8 or the groups



where p and r are 0 or 1 and are the same or different;

$N(R^8)_2$ may form a 5-7 membered ring such as pyrrolidine, piperidine or morpholine;

R^{11} is OH, OC_{1-4} alkyl or $N(R^8)_2$ (where R^8 may be the same or different); and

R^{12} is H, COR^8, CO_2R^8 (where R^8 is not H), $CONHR^8$ or SO_2R^8 (where R^8 is not H);

and the salts, solvates and hydrates thereof.

Description of the Invention

It will be appreciated that the compounds according to the invention can contain one or more asymmetrically substituted carbon atoms, for example those marked with an asterisk in formula (I). The presence of one or more of these asymmetric centres in a compound of formula (I) can give rise to stereoisomers, and in each case the invention is to be understood to extend to all such stereoisomers, including enantiomers and diastereomers, and mixtures including racemic mixtures thereof.

In the formulae herein, the ~ line is used at a potential asymmetric centre to represent the possibility of *R*- and *S*- configurations, the < line and the ..... line to represent a unique configuration at an asymmetric centre.

As used in this specification, alone or in combination, the term "C_{1-7} alkyl" refers to a straight or branched chain alkyl moiety having from one to seven carbon atoms, including for example, methyl, ethyl, propyl, isopropyl, butyl, *t*-butyl, pentyl, hexyl, heptyl and the like.

The term "C_{1-6} alkyl" refers to straight or branched chain alkyl moiety having from one to six carbon atoms, including for example, methyl, ethyl, propyl, isopropyl, butyl, *t*-butyl, pentyl, hexyl and the like.

The term "C_{1-4} alkyl" refers to a straight or branched chain alkyl moiety having from one to four carbon atoms, including for example, methyl, ethyl, propyl, isopropyl, butyl, *t*-butyl and the like.

The term "C_{2-6} alkenyl" refers to a straight or branched chain alkyl moiety having two to six carbon atoms and having in addition one double bond, of either E or Z stereochemistry where applicable. This term would include for example, vinyl, 1-propenyl, 1- and 2-butenyl, 2-methyl-2-propenyl etc.

The term "cyclo(C_{3-6})alkyl" refers to a saturated alicyclic moiety having from three to six carbon atoms and includes for example cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl and the like.

The term "cyclo(C_{4-6})alkenyl" refers to an alicyclic moiety having from four to six carbon atoms and having in addition one double bond. This term would include for example cyclopentenyl or cyclohexenyl.

There term "aryl" means an optionally substituted phenyl or naphthyl group with the substituent(s) being selected, for example, from halogen, trifluoromethyl, C_{1-6} alkyl, alkoxy, phenyl and the like.

The term "heteroaryl" refers to aromatic ring systems of five to ten atoms of which at least one atom is selected from O, N and S.

The term "halogen" means fluorine, chlorine, bromine or iodine.

The terms "protected amino" and "protected carboxy" mean amino and carboxy groups which are protected in a manner familiar to those skilled in the art. For example, an amino group can be protected by a benzyloxycarbonyl, *tert*-butoxycarbonyl, acetyl or like groups, or in the form of a phthalimido or like group. A carboxyl group can be protected in the form of a readily cleavable ester such as the methyl, ethyl, benzyl or *tert*-butyl ester.

Salts of compounds of formula (I) include pharmaceutically acceptable salts, for example acid addition salts derived from inorganic or organic acids, such as hydrochlorides, hydrobromides, p-toluenesulphonates, phosphates, sulphates, perchlorates, acetates, trifluoroacetates, propionates, citrates, malonates, succinates, lactates, oxalates, tartrates and benzoates.

Salts may also be formed with bases. Such salts include salts derived from inorganic or organic bases, for example alkali metal salts such as magnesium or calcium salts, and organic amine salts such as morpholine, piperidine, dimethylamine or diethylamine salts.

When the "protected carboxy" group in compounds of the invention is an esterified carboxyl group, it may be a metabolically labile ester of formula CO_2R^{11} where R^{11} may be an ethyl, benzyl, phenethyl, phenylpropyl, α- or β-naphthyl, 2,4-dimethylphenyl, 4-*tert*-butylphenyl, 2,2,2-trifluoroethyl, 1-(benzyloxy)benzyl, 1-(benzyloxy)ethyl, 2-methyl-1-propionyloxypropyl, 2,4,6-trmethylbenzyloxymethyl or pivaloyloxymethyl group.

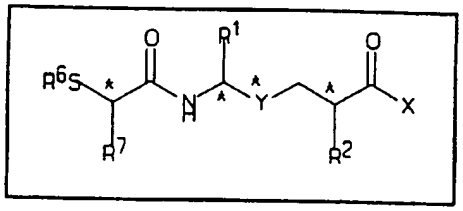
Compounds of the general formula (I) may be prepared by any suitable method known in the art and/or by the following processes, which itself forms part of the invention.

According to a second aspect of the invention, there is provided a process for preparing a compound of general formula (I) as defined above. It will be appreciated that where a particular stereoisomer of formula (I) is required, the synthetic processes described herein may be used with the appropriate homochiral starting material and/or isomers maybe resolved from mixtures using conventional separation techniques (e.g. HPLC).

The compounds according to the invention may be prepared by the following process. In the description and formulae below the groups R^1, R^2, R^3, R^4, R^5, R^6, R^7, R^8, R^9, R^{10}, R^{11}, R^{12}, R^{13},A,X and Y are as defined above, except where otherwise indicated. It will be appreciated that functional groups, such as amino, hydroxyl or carboxyl groups, present in the various compounds decribed below, and which it is desired to retain, may need to be in protected form before any reaction is initiated. In such instances, removal of the protecting group may be the final step in a particular reaction. Suitable protecting groups for such functionality will be apparent to those skilled in the art. For specific details see "Protective Groups in Organic Synthesis", Wiley Interscience, T W Greene, PGM Wuts.

A process for preparing compounds of general formula (I) comprises deprotecting (for example by hydrolysis) a compound of general formula (II)

(II)

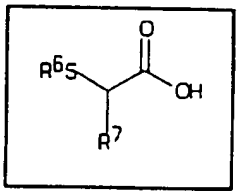


wherein R^6 represents a suitable protecting group (eg *tert*-butyl or acetate).

It will be appreciated that where a particular stereoisomer of formula (I) is required, this may be obtained by conventional resolution techniques such as high performance liquid chromatography. Where desired, however, appropriate homochiral starting materials may be used in the coupling reaction to yield a particular stereoisomer of formula (I). This is exemplified below.

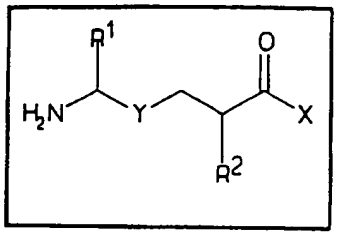
Intermediates of general formula (II) may be prepared by coupling an acid of formula (III)

(III)



wherein R^6 and R^7 are described previously, or an activated derivative thereof, with an amine of general formula (IV).

(IV)



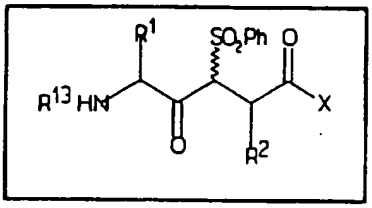
where R^1, R^2, X and Y are previously described.

Active derivatives of acids of formula (III) include for example acid anhydrides or acid halides, such as acid chlorides.

The coupling reaction may be performed using standard conditions for amination reactions of this type. Thus, the reaction may be achieved in a solvent, for example an inert organic solvent such as an ether, e.g. a cyclic ether such as tetrahydrofuran, an amide e.g. a substituted amide such as dimethylformamide, or a halogenated hydrocarbon such as dichloromethane at a low temperature e.g. -30°C to ambient temperature, such as -20°C to 0°C, optionally in the presence of as base, e.g. an organic base such as an amine, e.g. triethylamine or a cyclic amine such as *N*-methylmorpholine. Where an acid of formula (III) is used, the reaction may additionally be performed in the presence of a condensing agent, for example a diimide such as *N,N'*-dicyclohexylcarbodiimide, advantageously in the presence of a triazole such as 1-hydroxybenzotriazole. Alternatively, the acid may be reacted with a chloroformate for example ethylchloroformate, prior to reaction with the amine of formula (IV).

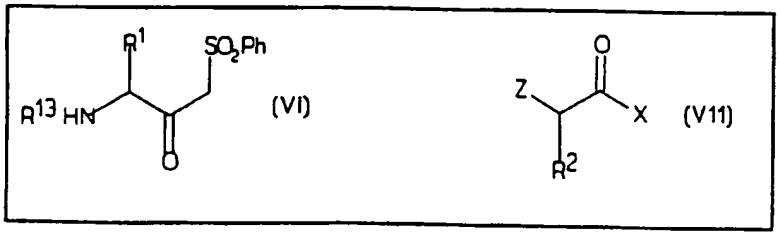
When Y=CO, amines of general formula (IV) may be prepared by desulphonylation of a sulphone of general formula (V)

(V)



wherein R^{13} is a suitable protecting group, such as *tert*-butyloxycarbonyl or benzyloxycarbonyl, followed by removal of any such protecting groups.

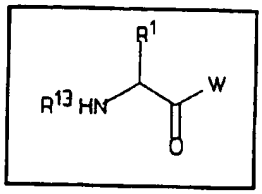
Amines of formula (V) may be prepared by alkylation of a sulphone of general formula (VI) with an alkylating agent of formula (VII)



wherein Z is a suitable leaving group such as a halide (eg chloride, bromide etc) or a sulphonate ester (eg methanesulphonate).

Sulphones of formula (VI) may be prepared by the reaction of a methylphenylsulphone anion with a suitably protected α-amino acid derivative of general formula (VIII)

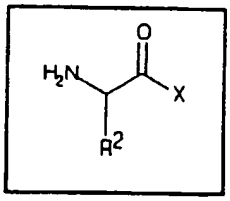
(VIII)



Derivatives of (VIII) include esters (ie W=OMe), amides [ie W=NMe(OMe)], acyl halides (ie W=Cl) or anhydrides (ie W=OCO_2Me).

Alkylating agents of formula (VII) maybe prepared from α-amino acid derivatives of general formula (IX)

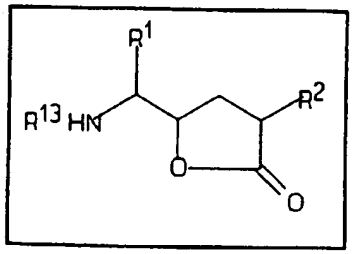
(IX)



α-Thioacetic acids and amino acids and their derivatives such as depicted by general formulae (III) and (VIII) or (IX) respectively can be obtained in optically pure or racemic form. In the homochiral form they provide asymmetric building blocks for the enantiospecific synthesis of compounds of general formula (I). Many of these derivatives can be readily obtained from commercially available starting materials using methods known to those skilled in the art. (See "The Practice of Peptide Synthesis" by M. Bodanszk *et al*, Springer Verlag, New York, 1984, P. L. Durette, WO92/21360).

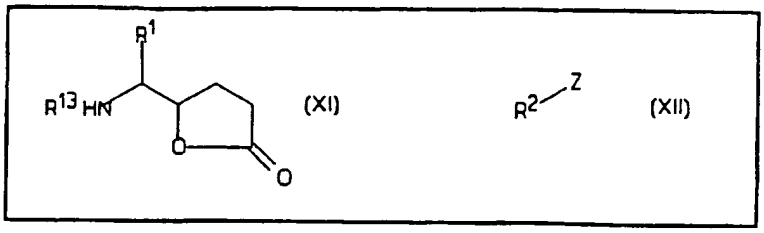
When Y=CHOH, amines of formula (IV) may be prepared by nucleophilic ring opening of a lactone of general formula (X) with a nucleophile derived from X as previously described

(X)



where R^1, R^2 and R^{13} are described previously.

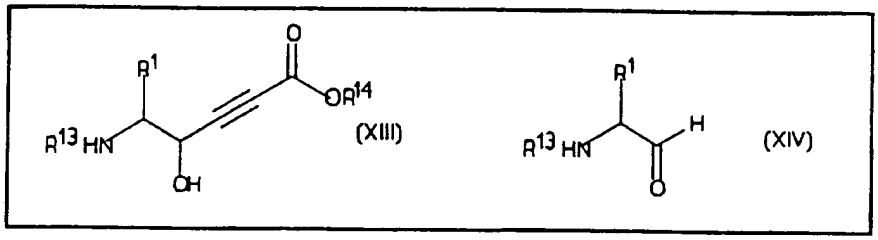
Lactones of general formula (X) may be prepared by alkylation of lactones of general formula (XI) with alkylating agents of general formula (XII)



wherein Z has been described previously.

Alkylating agents of formula (XII) are often commercially available or can be readily obtained from commercially available starting materials using methods known to those skilled in the art.

Lactones of formula (XI) may be prepared by reduction/cyclisation of acetylenes of general formula (XIII), where R^{14} is a C_{1-4} alkyl group (eg methyl, ethyl etc), which are in turn available by acetylide addition to an aldehyde of general formula (XIV) according to A. H. Fray, *J. Org. Chem.*, 1986, 51, 4828.



Aldehydes of formula (XIV) may be prepared by reduction of a suitably protected a-amino acid derivative of general formula (VIII) as described previously.

Compounds of formula (I) may also be prepared by interconversion of other compounds of formula (I). Thus, for example, a compound of formula (I) wherein R^1 is a C_{1-6} alkyl group may be prepared by hydrogenation (using palladium on carbon in suitable solvent, such as an alcohol - eg ethanol) of a compound of formula (I) wherein R^1 is a C_{2-6} alkenyl group. A further example would include a compound of formula (I) where Y is CHOH which may be prepared by reduction of a compound of formula (I) wherein Y is CO, conversely, a compound of formula (I) where Y is CO may be prepared by oxidation of a compound of formula (I) wherein Y is CHOH

Any mixtures of final products or intermediates obtained can be separated on the basis of the pysico-chemical differences of the constituents, in known manner, into the pure final products or intermediates, for example by chromatography, distillation, fractional crystallization, or by formation of a salt if appropriate or possible under the circumstances.

The compounds according to the invention exhibit *in vitro* inhibiting activities with respect to stromelysin, collagenase and gelatinase. Compounds according to the invention also exhibit *in vitro* inhibition of TNF release. The activity and selectivity of the compounds may be determined by use of the appropriate enzyme inhibition test, for example as described in Example A below and in WO-A-9611209 etc which also give other tests (Examples B to G) appropriate for testing compounds of the invention.

This invention also relates to a method of treatment for patients (including man and/or mammalian animals raised in the dairy, meat or fur industries or as pets) suffering from disorders or diseases which can be attributed to stromelysin as previously described, and more specifically, a method of treatment involving the administration of the matrix metalloproteinase inhibitors of formula (I) as the active constituents.

Accordingly, the compounds of formula (I) can be used among other things in the treatment of osteoarthritis and rheumatoid arthritis, and in diseases and indications resulting from the over-expression of these matrix metalloproteinases such as found in certain metastatic tumour cell lines.

As mentioned above, compounds of formula (I) are useful in human or veterinary medicine since they are active as inhibitors of TNF and MMPs. Accordingly in another aspect, this invention concerns:

a method of management (by which is meant treatment of prophylaxis) of disease or conditions mediated by TNF and/or MMPs in mammals, in particular in humans, which method comprises administering to the mammal an effective, amount of a compound of formula (I) above, or a pharmaceutically acceptable salt thereof; and

a compound of formula (I) for use in human or veterinary medicine, particularly in the management (by which is meant treatment or prophylaxis) of diseases or conditions mediated by TNF and/or MMPs; and

the use of a compound of formula (I) in the preparation of an agent for the management (by which is meant treatment or prophylaxis) of diseases or conditions mediated by TNF and/or MMPs.

The disease or conditions referred to above include inflammation, fever, cardiovascular effects, haemorrhage, coagulation and acute phase response, cachexia and anorexia, acute infections, shock states, graft versus host reactions and autoimmune disease; and those involving tissue breakdown such as bone resportion, inflammatory diseases, dermatological conditions, tumour growth, angiogenesis and invasion by secondary metastases, in particular rheumatoid arthritis, osteoarthritis, periodontitis, gingivitis, corneal ulceration, tumour growth, angiogenesis and invasion by secondary metastases.

For the treatment of rheumatoid arthritis, osteoarthritis, and in diseases and indications resulting from the over-expression of matrix metalloendoproteinases such as found in certain metastatic tumour cell lines or other diseases mediated by the matrix metalloendoproteinases or increased TNF production, the compunds of formula (I) may be administered orally, topically, parenterally, by inhalation spray or rectally in dosage unit formulations containing non-toxic pharmaceutically acceptable carriers, adjuvants and vehicles. The term parenteral as used herein includes subcutaneous injections, intravenous, intramuscular, intrasternal injection or infusion techniques. In addition to the treatment of warm-blooded animals such as mice, rats, horses, cattle, sheep, dogs, cats etc, the compounds of the invention are effective in the treatment of humans.

The pharmaceutical composition containing the active ingredient may be in a form suitable for oral use, for example, as tablets, troches, lozenges, aqueous or oily suspensions, dispersible powders or granules, emulsions, hard or soft capsules, or syrups or elixirs. Compositions intended for oral use may be prepared according to any method known to the art for the manufacture of pharmaceutical compositions and such compositions may contain one or more agents selected from the group consisting of sweetening agents, flavouring agents, colouring agents and preserving agents in order

to provide pharmaceutically elegant and palatable preparations. Tablets contain the active ingredient in admixture with non-toxic pharmaceutically acceptable excipients which are suitable for the manufacture of tablets. These excipients may be for example, inert diluents, such as calcium carbonate, sodium carbonate, lactose, calcium phosphate or sodium phosphate; granulating and disintegrating agents, for example corn starch, or alginic acid; binding agents, for example starch, gelatin or acacia, and lubricating agents, for example magnesium stearate, stearic acid or talc. The tablets may be uncoated or they may be coated by known techniques to delay disintegration and absorption in the gastointestinal tract and thereby provide a sustained action over a longer period. For example, a time delay material such as glyceryl monostearate or glyeryl distearate may be employed. They may also be coated by the techniques described in the US Patents 4,256,108;4,166,452; and 4,265,874 to form osmotic therapeutic tablets for control release.

Formulations for oral use may also be presented as hard gelatin capsules where in the active ingredient is mixed with an inert solid diluent, for example calcium carbonate, calcium phosphate or kaolin, or as soft gelatin capsules wherein the active ingredient is mixed with water or an oil medium, for example peanut oil, liquid paraffin or olive oil.

Aqueous suspensions contain the active materials in admixture with excipients suitable for the manufacture of aqueous suspensions. Such excipients are suspending agents, for example sodium carboxymethylcellulose, methylcellulose, hydroxy-propylmethylcellulose, sodium alginate polyvinyl-pyrrolidone, gum tragacanth and gum acacia; dispersing or wetting agents may be a naturally occuring phosphatide, for example lecithin, or condensation products of an alkylene oxide with fatty acids, for example polyoxyethylene stearate, or condensation products of ethylene oxide with long chain aliphatic alcohols, for example heptadecaethyleneoxycetanol, or condensation products of ethylene oxide with partial esters dervied from fatty acids and a hexitol such a polyoxyethylene with partial esters derived from fatty acids and hexitol anhydrides, for example polyoxyethylene sorbitan monooleate. The aqueous suspensions may also contain one or more preservatives, for example ethyl, or n-propyl, p-hydroxybenzoate, one or more colouring agents, one or more flavouring agents, and one or more sweetening agents, such as sucrose or saccharin.

Oily suspensions may be formulated by suspending the active ingredient in a vegetable oil, for example arachis oil, olive oil, sesame oil or coconut oil, or in a mineral oil such as liquid paraffin. The oily suspensions may contain a thickening agent, for example beeswax, hard paraffin or cetyl alcohol. Sweetening agents such as those set forth above, and flavouring agents may be added to provide a palatable oral preparation. These compositions may be preserved by the addition of an anti-oxidant such as ascorbic acid.

Dispersible powders and granules suitable for preparation of an aqueous suspension by the addition of water provide the active ingredient in admixture with a dispersing or wetting agent, suspending agent and one or more preservatives. Suitable dispersing or wetting agents and suspending agents are exemplified, for example sweetening, flavouring and colouring agents, may also be present.

The pharmaceutical compositions of the invention may also be in the form of oil-in-water emulsions. The oily phase may be a vegetable oil, for example olive oil or arachis oil, or a mineral oil, for example liquid paraffin or mixtures of these. Suitable emulsifying agents may be naturally-occuring gums, for example gum acacia or gum tragacanth, naturally-occuring phosphatides, for example soya bean, lecithin, and esters or partial esters derived from fatty acids and hexitol anhydrides, for example sorbitan monooleate and condensation products of the said partial esters with ethylene oxide, for example polyoxyethylene sorbitan monooleate. The emulsions may also contain sweetening and flavouring agents.

Syrups and elixirs may be formulated with sweetening agents, for example gycerol, propylene glycol, sorbitol or sucrose. Such formulations may also contain a demulcent, a preservative and flavouring and colouring agents. The pharmaceutical compositions may b in the form of a sterile injectable aqueous or oleagenous suspension. This suspension may be formulated according to the known art using those suitable dispersing or wetting agents and suspending agents which have been mentioned above. The sterile injectable preparation may also be in a sterile injectable solution or suspension in a non-toxic parenterally-acceptable diluent or solvent, for example as a solution in 1,3-butane diol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose any bland fixed oil may be employed including synthetic mono- or diglycerides. In addition, fatty acids such as oleic acid find use in the preparation of injectables.

The compounds of formula (I) may also be administered in the form of suppositories for rectal administration of the drug. These compositions can be prepared by mixing the drug with a suitable non-irritating excipient which is solid at ordinary temperatures but liquid at the rectal temperature and will therefore melt in the rectum to release the drug. Such materials are cocoa butter and polyethylene glycols.

For topical use, creams, ointments, jellies, solutions or suspensions, etc containing the compounds of Formula (I) are employed. (For purposes of this application, topical application shall include mouth washes and gargles.)

Dosage levels of the order of from about 0.05 mg to about 140 mg per kilogram of body weight per day are useful in the treatment of the above-indicated conditions (about 2.5 mg to about 7 gms per patient per day). For example, inflammation may be effectively treated by the administration of from about 0.01 to 50 mg of the compound per kilogram of body weight per day (about 0.5 mg to about 3.5 gms per patient per day).

The amount of active ingredient that may be combined with the carrier materials to produce a single dosage form will vary depending upon the host treated and the particular mode of administration. For example, a formulation intended for the oral administration of humans may vary from about 5 to about 95 percent of the total composition. Dosage unit forms will generally contain between from about 1 mg to about 500 mg of an active ingredient.

It will be understood, however, that the specific dose level for any particular patient will depend upon a variety of factors including the activity of the specific compound employed, the age, body weight, general health, sex, diet time of administration, route of administration, rate of excretion, drug combination and the severity of the particular disease undergoing therapy.

The following non-limiting Example is intended to illustrate the preparation of compounds of Formula (I), and as such is not intended to limit the invention as set forth in the claims.

In the Examples, the following abbreviations are used:

RT Room temperature
EDC 1-(3-Dimethylaminopropyl)-3-ethyl carbodiimide, hydrochloride
TNFα Tumour necrosis factor α
ELISA Enzyme linked immunosorbent assay
LPS Lipopolysaccharide (Endotoxin)

Intermediate 1 **(5*S*)-5-[(1*S*)-1-[*N*-[(1,1-dimethylethoxy)carbonyl]amino]-3-methylbutyl]dihydrofuran-2(3*H*)-one**

The compound was prepared according to the procedure of Kleinman *et al* [*J. Org. Chem.* 1986, *51*, 4828]

Intermediate 2 **(3*S*, 5*S*)-3-Phenylmethyl-5-[(1*S*)-1-[*N*-[(1,1-dimethylethoxy)carbonyl]amino]-3-methylbutyl]dihydrofuran-2(3*H*)-one**

n-Butyllithium (1.6M in hexanes, 2.6 ml, 4.2 mmol) was added to a solution of hexamethyldisilazane (0.90 ml, 4.25 mmol) in THF (5 ml) at 0°C and the pale yellow solution was stirred for 15 min, then cooled to - 78 °C. A solution of intermediate 1 (0.52 g, 1.92 mmol) in THF (5 ml) was added dropwise over 15 min, the mixture was stirred for 30 min, then benzyl bromide (0.25 ml, 2.1 mmol) was added dropwise. The solution was stirred at -78 °C for 60 min, then warmed to - 40 °C and stirred for a further 90 min. Saturated ammonium chloride solution (30 ml) was added and the mixture extracted with ether. The solvent was washed with brine, dried and evaporated and the residue purified by flash column chromatography (3:1 hexanes:ether) to give the title compound as a colourless solid (0.18 g, 26 %).

TLC R_f 0.25 (3:1 hexanes:ether).

Intermediate 3 **(2*S*, 4*S*, 5*S*)-*N*-Methyl 2-benzyl-4-hydroxy-5-[[(1,1-dimethylethoxy)-carbonyl]amino]-7-methyloctanamide**

A solution of intermediate 2 (0.18 g, 0.52 mmol) was heated at 70 °C in a mixture of THF (10 ml) and aqueous methylamine (40 %, 5 ml) for 2 h. The solution was cooled , diluted with EtOAc (100 ml) and washed with water and brine. The solvent was dried and evaporated to give a colourless solid which was purified by flash column chromatography (1:1 CH_2Cl_2:EtOAc) to give the title compound as acolourless solid (145 mg, 71 %).

TLC R_f 0.35 (1:1 CH_2Cl_2:EtOAc).

Example 1 **(2*S*, 4*S*, 5*S*, 2'*S*)-*N*-methyl-2-benzyl-4-hydroxy-5-[*N*-[(2'-acetylthio)-5'-phthalimidopentanoyl]amino]-7-methyloctanamide**

A solution of intermediate 3 (145 mg) was dissolved in a mixture of TFA (5 ml) and CH_2Cl_2 and the solution stirred for 2 h. The mixture was evaporated and the residue dissolved in ether and extracted with water. The aqueous phase was neutralised with $NaHCO_3$ and extracted with CH_2Cl_2. The solvent was dried and evaporated and the residue dissolved in dry THF (5 ml). A solution of 2-(acetylthio)-5-phthalimidopentanoic acid (100 mg, 0.3 mmol) was added, followed by HOBT (46 mg) and EDC (65 mg) and the mixture was stirred at RT overnight. CH_2Cl_2 (50 ml) was added and the solution was washed with 0.5 M HCl, sat. $NaHCO_3$ and brine, dried and evaporated. The residue was purified by flash column chromatography (EtOAc) to give the title compound as acolourless solid (26 mg).

TLC R_f 0.41 (EtOAc)

Example A

Collagenase inhibition activity

The potency of compounds of general formula (I) to act as inhibitors of collagenase was determined by the procedure of Cawston and Barrett (Anal. Biochem., 99:340-345, 1979) whereby a 1mM solution of th inhibitor being tested or dilutions thereof was incubated at 37°C for 16 hours with collagen and collagenase (buffered with 50 mM Tris, pH 7.6 containing 5 mM $CaCl_2$, 0.05% Brij 35, 60 mM NaCl and 0.02% NaN_3). The collagen was acetylated ^3H or ^{14}C-collagen prepared by the method of Cawston and Murphy (Methods in Enzymology, 80:711, 1981). The choice of radiolabel did not alter the ability of collagenase to degrade the collagen substrate. The samples were centrifuged to sediment undigested collagen and an aliquot of the radioactive supernatant removed for assay on a scintillation counter as a measure of hydrolysis. The collagenase activity in the presence of 1mM inhibitor, or a dilution thereof, was compared to activity in a control devoid of inhibitor and the results reported as that inhibitor concentration effecting 50% inhibition of the collagenase (IC_{50}).

Example B

Stromelysin inhibition activity

The potency of compounds of general formula (I) to act as inhibitors of stromelysin was determined using the procedure of Nagase *et al* (Methods in Enzymology Vol 254, 1994), whereby a 0.1 mM solution of the inhibitor being tested or dilutions thereof was incubated at 37°C for 16 hours with stromelysin and ^3H transferrin (buffered with 50 mM Tris, pH 7.6 containing 10 mM $CaCl_2$, 150M NaCl, 0.05% Brij, 35, and 0.02% NaN_3). The transferrin was carboxymethylated with ^3H iodoacetic acid. The stromelysin activity in the presence of 1 mM, or a dilution thereof, was compared to activity in a control devoid of inhibitor and the results reported as that inhibitor concentration effecting 50% inhibition of the stromelysin (IC_{50})

Example C

Gelatinase inhibition activity

The potency of the compounds of general formula (I) to act as inhibitors of gelatinase was determined using the procedure of Harris & Krane (Biochem Biophys. Acta, 258:566 - 576, 1972), whereby a 1 mM solution of the inhibitor being tested or dilutions thereof was incubated at 37°C for 16 hours with gelatinase and heat denatured ^3H or ^{14}C-acetylated collagen (buffered with 50 mM Tris, pH 7.6 containing 5 mM $CaCl_2$, 0.05% Brij 35 and 0.02% NaN_3). The ^3H or ^{14}C gelatin was prepared by denaturing ^3H or ^{14}C-collagen produced according to the method of Cawston and Murphy (Methods in Enzymology, 80:711, 1981) by incubation at 60°C for 30 minutes. Undigested gelatin was precipitated by addition of trichloroacetic acid and centrifugation. The gelatinase activity in the presence of 1 mM, or dilution thereof, was compared to the activity in a control devoid of inhibitor and results reported as that inhibitor concentration effecting 50% inhibition of the gelatinase (IC_{50}).

Example D

MMP Inhibition Activity-Fluorimetric Assay

The potency of compounds of general formula (I) to act as inhibitors of collagenase-1 (MMP- 1), collagenase-2 (MMP-8), gelatinase-A (MMP-2), gelatinase-B (MMP-9) and stromelysin-1 (MMP-3) was determined using the following procedure:

Inhibitors are dissolved in dimethylsulphoxide containing 0.02% β-mercaptoethanol and serial dilutions are prepared. Activated enzyme is incubated in assay buffer containing 50 mM Tris, pH 7.4, 5 mM $CaCl_2$, 0.002% NaN_3 and Brij 35 in the presence and absence of inhibitor. Samples are

preincubated at 37°C for 15 minutes before the addition of the fluorimetric substrate (McaPro-Leu-Dpa-Ala-Arg-NH_2) to a final concentration of 10 μM. The assay is incubated for 90 minutes at 37°C and then read in a Fluoroscan II at γ_{ex} (355 nm) and γ_{em} (460 nm).

The enzyme activity was compared to activity in a control devoid of inhibitor and the results reported as that inhibitor concentration effecting 50% inhibition of the stromelysin (IC_{50}).

Example E

Inhibition of TNFα production

The potency of the compounds of general formula (I) to act as inhibitors of the production of TNFα was determined using the following procedure. A 1 mM solution of the inhibitor being tested or dilutions thereof was incubated at 37°C in an atmosphere of 5% CO_2 with THP-1 cells (human monocytes) suspended in RPMI 1640 medium and 20 μM β-mercaptoethanol at a cell density of 1 x 10^6/ml and stimulated with 5 μg/ml final concentration of LPS. After 18 hours the supernatant is assayed for the levels of TNFα using a commercially available ELISA kit (R & D Systems).

The activity in the presence of 0.1 mM inhibitor or dilutions thereof was compared to activity in a control devoid of inhibitor and results reported as that inhibitor concentration effecting 50% inhibition of the production of TNFα.

Example F

Adjuvant arthritic rat model

Compounds of general formula (I) were evaluated in an adjuvant arthritis model in the rat based on the methods employed by B.B. Newbould (1963), Br.J.Pharmacol, 21, 127-136 and C.M. Pearson and F.D. Wood (1959), Arthritis Rheum, 2, 440-459. Briefly male Wistar rats (180-200 g) were injected at the base of the tail with Freund's adjuvant. Twelve days later the responding animals were randomised into experimental groups. Compounds of general formula (I) were dosed either orally as a suspension in 1% methylcellulose or intraperitoneally in 0.2% carboxymethylcellulose from day 12 to the end of the experiment on day 22. Hind paw volumes were measured every two days from day 12 onwards and X-rays were taken of the hind feet on completion of the experiment. Results were expressed as the percent increase of foot volume over day 12 values.

Example G

Mouse ovarian carcinoma xenograft model

Compounds of general formula (I) were evaluated in an ovarian carcinoma xenograft model of cancer, based on that described by B. Davies *et al* (1993), Cancer Research, 53, 2087-2091. This model, in brief, consists of inoculating female nu/nu mice with 1 x 10^9 OVCAR3-icr cells into the peritoneal cavity. Compounds of general formula (I) are administered by the oral route as a suspension in 1% methylcellulose or intraperitoneally as a suspension in phosphate buffered saline in 0.01% Tween-20. At the conclusion of the experiment (4-5 weeks) the number of peritoneal cells are counted and any solid tumour deposits weighed. In some experiments tumour development is monitored by measurement of tumour specific antigens.

Example H

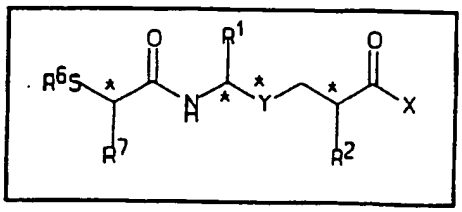
Rat mammary carcinoma model

Compounds of general formula (I) were evaluated in a HOSP.1 rat mammary carcinoma model of cancer (S.Eccles *et al* (1995), Cancer Research, in press). This model consists of the intravenous inoculation of female CBH/cbi rats with 2 x 10^4 tumour cells into the jugular vein. Compounds of general formula (I) are administered by the oral route as a suspension in 1% methylcellulose or intraperitoneally as a suspension in phosphate buffered saline in 0.01% Tween-20. At the conclusion of the experiment (4-5 weeks) the animals were killed, the lungs were removed and individual tumours counted after 20 hours fixation in Methacarn.

CLAIMS

1. Compound of general formula (I)

(I)



wherein:

R^1 represents a C_{1-7} alkyl, C_{2-6} alkenyl, (C_{1-6} alkyl)aryl, (C_{1-6} alkyl)heteroaryl or C_{1-6} alkyl-AR^8 group where A is O, NR^8 or $S(O)_n$ where n = 0-2, and R^8 is H, C_{1-4} alkyl, aryl, heteroaryl, (C_{1-4} alkyl)aryl or (C_{1-4} alkyl)heteroaryl; if A = NR^8 the groups R^8 may be the same or different.

R^2 represents a $[Alk]_mR^5$ group where Alk is a C_{1-6} alkyl or C_{2-6} alkenyl group and m = 0-1;

X is NR^3R^4 where R^3 is hydrogen or the group C_{1-6} alkyl optionally substituted by amino (NH_2), aryl, arylamino, protected amino, di (C_{1-6}alkyl)amino, mono (C_{1-6}alkyl)amino, CO_2H, protected carboxyl, carbamoyl, mono (C_{1-6}alkyl) carbamoyl, di (C_{1-6}alkyl) carbamoyl, and R^4 is hydrogen or a C_{1-6} alkyl group; NR^3R^4 may also form a 5-7 membered ring such as pyrrolidine, piperidine or morpholine;

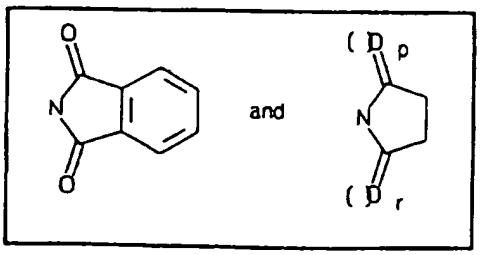
Y represents CHOH, $CHNH_2$ or C=O;

R^5 represents an optionally substituted cyclo(C_{3-6})alkyl, cyclo(C_{3-6})alkenyl, C_{1-6} alkyl, aryl, heteroaryl, CO_2R^8, $CONHR^8$, $NHCO_2R^8$, $NHSO_2R^8$, $NHCOR^8$ or AR^8 group;

R^6 represents H or R^9CO where R^9 is C_{1-4} alkyl or aryl;

R^7 represents aryl (optionally substituted with R^{10}), heteroaryl (optionally substituted with R^{10}), C_{1-4} alkyl (optionally substituted with R^{10}), C_{1-4} alkenyl (optionally substituted with R^{10}), (C_{1-4} alkyl)aryl (optionally substituted with R^{10}), (C_{1-4} alkyl)heteroaryl (optionally substituted with R^{10}), cyclo(C_{3-6})alkyl (optionally substituted with R^{10}) or cyclo(C_{3-6})alkenyl (optionally substituted with R^{10});

R^{10} represents H, SR^6, COR^{11}, $N(R^8)_2$ (where R^8 may be the same or different), NR^8R^{12}, OR^8, COR^8 or the groups;



where p and r are 0 or 1 and are the same or different;

$N(R^8)_2$ may form a 5-7 membered ring such as pyrrolidine, piperidine or morpholine;

R^{11} represents OH, OC_{1-4} alkyl or $N(R^8)_2$ (where R^8 may be the same or different); and

R^{12} represents H, COR^8, CO_2R^8 (where R^8 is not H), $CONHR^8$ or SO_2R^8 (where R^8 is not H);

and the salts, solvates and hydrates thereof.

2. A compound of claim 1, which is (2*S*, 4*S*, 5*S*, 2'*S*)-*N*-methyl-2-benzyl-4-hydroxy-5-[*N*-[(2'-acetylthio)-5'- phthalimidopentanoyl]amino]-7-methyloctanamide.

3. A compound of claim 1, in the form of a single enantiomer or diastereomer, or a mixture of such isomers.

4. A pharmaceutical composition for use in therapy, comprising a compound of any preceding claim, and a pharmaceutically-acceptable diluent or carrier.

5. Use of a compound of any of claims 1 to 3, for the manufacture of a medicament for the treatment or prevention of a condition associated with matrix metalloproteinases or that is mediated by TNFα or L-selectin sheddase.

6. Use according to claim 5, wherein the condition is selected from cancer, inflammation and inflammatory diseases, tissue degeneration, periodontal disease, ophthalmological disease, dermatological disorders, fever, cardiovascular effects, haemorrhage, coagulation and acute phase response, cachexia and anorexia, acute infection, HIV infection, shock states, graft versus host reactions, autoimmune disease, reperfusion injury, meningitis and migraine.

7. Use according to claim 5, wherein the condition is selected from tumour growth, angiogenesis, tumour invasion and spread, metastases, malignant ascites and malignant pleural effusion.

8. Use according to claim 5, wherein the condition is selected from rheumatoid arthritis, osteoarthritis, osteoporosis, asthma, multiple sclerosis, neurodegeneration, Alzheimer's disease, atherosclerosis, stroke, vasculitis, Crohn's disease and ulcerative colitis.

9. Use according to claim 5, wherein the condition is selected from corneal ulceration, retinopathy and surgical wound healing.

10. Use according to claim 5, wherein the condition is selected from psoriasis, atopic dermatitis, chronic ulcers and epidermolysis bullosa.

11. Use according to claim 5, wherein the condition is periodonititis or gingivitis.

12. Use according to claim 5, wherein the condition is selected from rhinitis, allergic conjunctivitis, eczema and anaphylaxis.

13. Use according to claim 5, wherein the condition is selected from ristinosis, congestive heart failure, endometriosis, atherosclerosis and endosclerosis.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/GB 96/02439

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C07D209/48 C07C323/60 C07C327/32 A61K31/415 A61K31/22 A61K31/16

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C07C C07D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 95 13289 A (CHIROSCIENCE) 18 May 1995 see the whole document	1,4,5

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
7 January 1997	13. 01. 97

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+ 31-70) 340-2040, Tx. 31 651 epo nl, Fax (+ 31-70) 340-3016	English, R

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 96/02439

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO-A-9513289	18-05-95	AU-A-	8113394	29-05-95
		EP-A-	0728144	28-08-96
		FI-A-	961976	09-05-96
		HU-A-	73799	30-09-96
		NO-A-	961888	09-05-96
		PL-A-	314300	02-09-96

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